#28 6/20/02

PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:
Sastry, et al.

Serial No.: 08/869,386

Filed: June 5, 1997

For: COMPOSITIONS AND METHODS FOR ELICITING AN IMMUNE RESPONSE

Group Art Unit: 1648

Examiner: B. Nelson

Atty. Dkt. No.: UTSC:538/HYL

DECLARATION OF PRAMOD N. NEHETE

I, Pramod N. Nehete, hereby declare as follows:

1. I am an inventor of the subject matter disclosed and claimed in the referenced patent application. I am currently employed as an Assistant Professor in the Department of Veterinary Sciences at The University of Texas M. D. Anderson Cancer Center. I have expertise in the design, development and implementation of peptide-based vaccines for the treatment of viral infections, in particular HIV infections, as evidenced by my curriculum vitae, a copy of which is attached as Exhibit 1.

2. I understand that the examiner in charge of the referenced patent prosecution matter has taken the position that it has not been adequately demonstrated that the claimed R15K peptide would likely have clinical utility in the treatment of HIV. I am submitting this declaration to provide new information that clearly demonstrates the clinical utility of peptides incorporating the R15K sequence in the treatment of HIV. In this regard, I understand that my colleague and joint inventor, Dr. Jagan Sastry,

previously submitted a declaration providing the results of various *in vitro* studies carried out by our group using accepted in vitro systems for testing for treatments useful in HIV treatment. These studies are now published in Nehete *et al.* (Exhibit 2). In particular, the studies described in Dr. Sastry's earlier declaration and described in the Nehete *et al.* manuscript demonstrated that the central 15-21 amino acids in the V3 region of gp120 play an important role in HIV infection of $CD4^+$ cells. Peptides from this region bind to target host cells and inhibit the cellular entry of phenotypically distinct HIV-1 strains. Further, we showed that competition for peptide binding was observed with viral particles, but not with recombinant gp120, sCD4, β-chemokines or an antibody to CXCR-4.

3. The use of a synthetic peptide-based anti-HIV therapeutic has been validated by a recent report, Kilby *et al.* (Exhibit 3) which reports the results of a clinical trial in human HIV patients wherein potent suppression of HIV load in patients was observed following intravenous administration of T-20, a peptide corresponding to the oligomerization domain of the HIV-1 gp41 transmembrane protein. Subsequently, T-20 has also been successfully tested for subcutaneous administration in humans (unpublished studies).

4. Using the SHIV-rhesus model, we investigated the therapeutic potential of the V3 peptide R15K, exhibiting anti-HIV activity, by using the protocol described for the clinical trial with the T-20 peptide in Kilby *et al.* In this model, infection of rhesus macaques with a chimeric virus, SHIV consisting of SIV core and HIV envelope sequences leads to a productive infection followed by severe loss of CD4 cells and AIDS-related pathology. Thus, in the SHIV-rhesus model, the rhesus macaques are exposed to

acute infection by SHIV as opposed to the chronic HIV infection targeted in the human clinical trial using T-20 as the anti-HIV therapeutic reagent. The following is the description of the study protocol and results obtained:

A. Two groups of two monkeys each were selected for this study. One group, referred to as control, received daily intravenous infusions of sterile saline (1 ml/dose/animal) for a total of 15 days. The second group of monkeys received the R15K peptide daily by the intravenous route (60mg in 1 ml/dose/monkey). On day two of the infusions, all four monkeys were challenged with the pathogenic $SHIV_{ku2}$ by the intravenous dose (1 ml containing 1000 $TCID_{50}$). At several days post-challenge, blood samples were collected from each of the animals for determining the viral load by analyzing the plasma samples by using the real-time RT-PCR methodology.

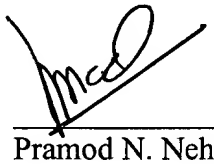
B. The data from the control and treated groups of animals is shown in the Fig.1 (Exhibit 4) as an average for the monkeys in each group. The time scale used is the number of days after viral infection, and therefore, the peptide treatment for the days prior to viral challenge is referred to as days with negative numbering. The differences in the viral loads between the control and treated groups of monkeys is shown both as RNA copy equivalents in either one micro-liter of the plasma (panel on the left) or 1 ml of the plasma after log-transformation (right-side panel). Further, the decrease in viral load in the R15K-treated monkeys is also shown in Fig.2 after log-transformation of the data in RNA copy equivalents.

C. It is clear that the viral load is reduced in the R15K-treated monkeys, and the decrease reached a maximum of 1.715 $\log_{10}$ units of RNA copy equivalents by day 19 post-challenge (Fig. 2) (Exhibit 4).

5. The SHIV-rhesus monkey model used in the foregoing studies, and one of the models that we have used to demonstrate efficacy of the R15K peptide in the treatment of HIV infections, is a model that is accepted by those of skill in the art of testing for HIV therapeutics. This model has been relied upon frequently in the scientific literature and shown to be a useful model for predicting efficacy of HIV vaccines. See, for exampe, the articles of I.M. Belyakov et al., H.L. Robinson et al., and D. H. Barouch et al., each attached as Exhibit 5 hereto. Using this accepted model, as shown above, the R15K peptide has been shown to be useful in significantly reducing viral load in infected individuals.

6. I hereby declare that all statements made herein of my knowledge are true and that all statements made herein on information and belief are believed to be true; and further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both, under §1001 of Title 18 of the U.S. Code, and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

6/17/02
Date



Pramod N. Nehete, Ph.D.